REMARKS

Claims 11, 12, 14-23 and 27-35 are pending in this application. Claims 1-10, 13 and 24-26 have been canceled without prejudice or disclaimer. Claims 34 and 35 have been newly added.

Claims 11, 14-23 and 28-33 have been amended for the sole reason of advancing prosecution. Applicants, by canceling or amending any claims herein, make no admission as to the validity of any rejection made by the Examiner against any of these claims. Applicants reserve the right to reassert any of the claims canceled herein or the original claim scope of any claim amended herein, in a continuing application.

Claim 11 has been amended to recite a "mutant lipase protein of *Candida antarctica* lipase B represented by SEQ.ID. No 14, wherein the #219 leucine is replaced by a hydrophilic amino acid selected from a group consisting of glutamine, histidine, arginine, lysine, serine, threonine, aspartic acid and glutamic acid." Support for claim 11, as amended, can be found throughout the specification and claims as originally filed, for example, in the specification at page 8, lines 2-5, page 10, lines 24-25 and page 11, lines 1-10.

Claim 14 has been amended to recite a "mutant lipase protein of *Candida antarctica* lipase B represented by SEQ.ID. No 14, wherein the #278 leucine is replaced by proline, and its amino acid sequence is represented by SEQ. ID. No 9." Support for claim 14, as amended, can be found throughout the specification and claims as originally filed, for example, in the specification at page 10, lines 18-23.

Claim 15 has been amended to recite a "mutant lipase protein of *Candida antarctica* lipase B represented by SEQ.ID. No 14, wherein the #219 leucine is replaced

by glutamine, and the #278 leucine is replaced by proline, and its amino acid sequence is represented by SEQ. ID. No 10." Support for claim 15, as amended, can be found throughout the specification and claims as originally filed, for example, in the specification at page 10, lines 24-25 and page 11, lines 1-10.

Claim 16 has been amended to recite a "polynucleotide encoding the mutant lipase protein of claim 11." Support for claim 16, as amended, can be found throughout the specification and claims as originally filed.

Claim 17 has been amended to recite the "polynucleotide as set forth in claim 16, wherein the nucleotide sequence is represented by SEQ. ID. No 8." Support for claim 17, as amended, can be found throughout the specification and claims as originally filed.

Claim 18 has been amended to recite a "polynucleotide encoding the mutant lipase protein of claim 14." Support for claim 18, as amended, can be found throughout the specification and claims as originally filed.

Claim 19 has been amended to recite a "polynucleotide, comprising a base sequence represented by SEQ. ID. No 7 coding the mutant lipase protein of claim 15." Support for claim 19, as amended, can be found throughout the specification and claims as originally filed.

Claim 20 has been amended to recite an "expression vector comprising the polynucleotide of claim 16." Support for claim 20, as amended, can be found throughout the specification and claims as originally filed.

Claim 21 has been amended to recite the "expression vector as set forth in claim 20, wherein the vector comprises a promoter gene, a secretion signal sequence gene, a

polynucleotide of SEQ. ID. No 8, a terminator gene and/or a surface display-mediating gene." Support for claim 21, as amended, can be found throughout the specification and claims as originally filed.

Claim 22 has been amended to recite an "expression vector comprising the polynucleotide of claim 18." Support for claim 22, as amended, can be found throughout the specification and claims as originally filed.

Claim 23 has been amended to recite an "expression vector comprising the polynucleotide of claim 19." Support for claim 23, as amended, can be found throughout the specification and claims as originally filed.

Claim 28 has been amended to recite a "transformant in which the expression vector of claim 22 is introduced." Support for claim 28, as amended, can be found throughout the specification and claims as originally filed.

Claim 29 has been amended to recite a "transformant in which the expression vector of claim 23 is introduced." Support for claim 29, as amended, can be found throughout the specification and claims as originally filed.

Claim 30 has been amended to recite a "method for producing the mutant lipase protein of claim 11, comprising cultivating the transformant of claim 27." Support for claim 30, as amended, can be found throughout the specification and claims as originally filed.

Claim 31 has been amended to recite a "method for producing the mutant lipase protein of claim 14, comprising cultivating the transformant of claim 28." Support for claim 31, as amended, can be found throughout the specification and claims as originally filed.

Claim 32 has been amended to recite a "method for producing the mutant lipase protein of claim 15, comprising cultivating the transformant of claim 29." Support for claim 32, as amended, can be found throughout the specification and claims as originally filed.

Claim 33 has been amended to recite the "method as set forth in any of claims 30 – 32, wherein the culture temperature is 2°C - 20°C lower than temperature of host cell culture." Support for claim 33, as amended, can be found throughout the specification and claims as originally filed.

New claims 34 and 35 have been added. New claim 34 recites the "method as set forth in any of claims 30 – 32, wherein the culture temperature is 25°C - 35°C and the transformant is *Hansenula.*" Support for new claim 34 can be found throughout the specification and claims as originally filed.

New claim 35 recites the "method as set forth in any of claims 30 – 32, wherein the culture temperature is 20°C - 28°C and the transformant is *Saccharomyces.*" Support for new claim 35 can be found throughout the specification and claims as originally filed.

No new matter has been added.

In view of the remarks set forth herein, further and favorable consideration is respectfully requested.

I. At page 2, of the Official Action, claims 1, 2, 4, 5 and 7-33 have been rejected under 35 USC § 112, second paragraph, as being indefinite.

The Examiner asserts that it is unclear whether a lipase gene is cloned into a vector already containing another lipase gene, or if the lipase gene is being introduced

into a vector without a lipase gene already present.

In view of the following, this rejection is respectfully traversed.

Claims 1, 2, 4, 5, 7-10, 13 and 24-26 have been canceled without prejudice or disclaimer rendering their rejection moot. Additionally, the rejection of claims 11, 12, 14-23, and 27-33 is respectfully traversed.

Applicants submit that amended claims 11, 12, 14 and 15 are directed to mutant lipases. Claims 16-19 are directed to the genes encoding the mutant lipases. Claims 20-23 are directed to the expression vectors containing these genes. Claims 27-29 are directed to the transformants in which the expression vectors are introduced. Claims 30-32 are directed to the methods for preparing the mutant lipases.

In view of the foregoing, it is submitted that claims 11, 12, 14-23 and 27-33 are clear and definite within the meaning of 35 USC § 112, second paragraph. Thus, the Examiner is respectfully requested to reconsider and withdraw this rejection.

II. At page 3, of the Official Action, claims 1, 3-6, 8 and 9, have been rejected under 35 USC § 103(a) as unpatentable over Svendsen et al. (US Patent No. 6,020,180) in view of Kim et al., "A cell surface display system using novel GPI-anchored proteins in Hansenula polymorpha," Yeast, vol. 19, pp. 1153-1163 (2002).

The Examiner asserts that Svendsen et al. describe a method for identifying lipase variants with improved properties. However, the Examiner acknowledges that Svendsen et al. do not teach a CWP1, GAS1, or TIP1 surface display mediating gene, or a surface display vector, or a MOX or GAPDH promoter. The Examiner asserts that Kim et al. describe a cell surface display system using anchored proteins in *Hansenula*. The Examiner also asserts that Kim et al. describe the cell surface display vector

includes a MOX or GAPDH promoter, an α-amylase signal sequence, and a CWP1, GAS1 or TIP1 surface display mediating gene, and that it would have been obvious to combine the teachings of Svendsen et al. with Kim et al. to arrive at the presently claimed subject matter.

Applicants respectfully submit that claims 1, 3-6, 8 and 9 have been canceled without prejudice or disclaimer, rendering this rejection moot. Thus, the Examiner is respectfully requested to reconsider and withdraw this rejection.

III. At page 3, of the Official Action, claim 2, has been rejected under 35 USC § 103(a) as unpatentable over Svendsen et al. in view of Kim et al. and in further view of Uppenberg et al., "The sequence, crystal structure determination and refinement of two crystal forms of lipase B from Candida Antarctica," Structure, vol. 2, pp. 293-308 (1994).

The Examiner asserts that it would have been obvious to combine the teachings of Svendsen et al. and Kim et al., with the use of *Candida antartica* lipase B cloned into a vector, transformed into *E. coli,* and then screened, as described by Uppenberg et al. to arrive at the presently claimed subject matter.

Applicants respectfully submit that claim 2 has been canceled without prejudice or disclaimer, rendering this rejection of claim 2 moot. Thus, the Examiner is respectfully requested to reconsider and withdraw this rejection.

IV. Amended Claims 11, 14-23 and 28-33.

Claims 11, 14-23 and 28-33 have been amended for the sole reason of advancing prosecution. No new matter has been added. Applicants submit that amended claims 11, 14-23 and 28-33 are novel and non-obvious. Amended claims 11, 12, 14 and 15 are directed to mutant lipases. Claims 16-19 are directed to the genes

encoding the mutant lipases. Claims 20-23 are directed to the expression vectors containing these genes. Claims 27-29 are directed to the transformants in which the expression vectors are introduced. Claims 30-32 are directed to the methods for preparing the mutant lipases.

Applicants submit that the mutant lipases of claims 12, 14 and 15 have specific amino acid sequences. In particular, claims 12, 14 and 15 are represented by SEQ. ID. Nos 9-11, respectively. The purified mutant lipases of claims 14 and 15 are represented by SEQ. ID. Nos 9 and 10, respectively. Additionally, Applicants submit that the purified mutant lipases of claims 12, 14 and 15 exhibited a 6-fold increase in enzymatic activities when compared to wild type lipases. *See* Examples 6 and 7, and Table 2 in the specification. Further, the mutant lipase of claims 12, represented by SEQ. ID No. 11, exhibited about a 3-fold increase in enzymatic activity when compared to wild type lipases. *See* Example 8, Figure 5 and Table 3 in the specification. Accordingly, the mutant lipases of claims 12-15 exhibited improved enzymatic activities over wild type lipases.

The mutant lipase of claim 11 is *Candida antarctica* lipase B, which has a hydrophilic amino acid, e.g. glutamine, histidine, arginine, lysine, serine, threonine, aspartic acid or glutamic acid, instead of leucine in the #219 position. The surface of *Candida antarctica* lipase B consists mainly of hydrophobic amino acids. However, by replacing the #219 leucine with a hydrophilic amino acid, the conformation of the lipase changes, accompanied by increases in enzymatic activity and stability in water. *See* the specification at page 11, lines 12-16, and page 28, lines 9-19.

Accordingly, Applicants respectfully request an indication that all of the pending

claims are now allowable.

V. New Claims 34-35.

Claims 34 and 35 have been newly added.

Applicants respectfully submit that new claims 34 and 35 are both novel and non-obvious, for at least the reason that new claims 34 and 35 depend from any of claims 30-32. Accordingly, Applicants respectfully request an indication that all of the pending claims are now allowable.

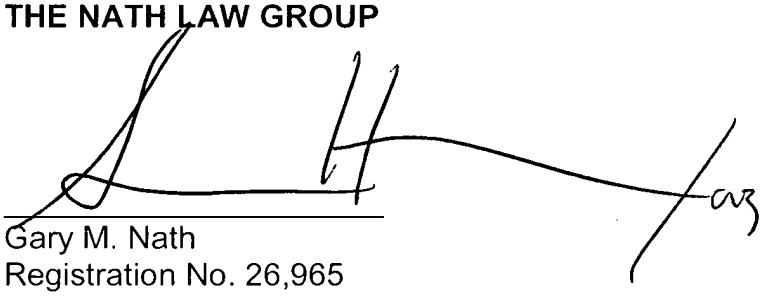
CONCLUSION

In view of the foregoing, Applicant submits that the pending claims are in condition for allowance. Early notice to that effect is earnestly solicited. The Examiner is invited to contact the undersigned attorney if it is believed that such contact will expedite the prosecution of the application.

In the event this paper is not timely filed, Applicant petitions for an appropriate extension of time. Please charge any fee deficiency or credit any overpayment to Deposit Account No. 14-0112.

Respectfully submitted,

THE NATH LAW GROUP



Gary M. Nath
Registration No. 26,965
Susanne M. Hopkins
Registration No. 33,247
Ari G. Zytcer
Registration No. 57,474
Customer No. 20529

Date: June 18, 2009
THE NATH LAW GROUP
112 South West Street
Alexandria, VA 22314
Tel: (703) 548-NATH
Fax: (703) 683-8396